CORRESPONDENCE

BNT162b2-Elicited Neutralization against New SARS-CoV-2 Spike Variants

TO THE EDITOR: Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) continues to evolve at a rapid pace, generating new variants that arouse concern. Variants that were first detected in California (B.1.429 lineage) and New York (B.1.526 lineage) are causing concern in the United States. A variant that was first detected in the United Kingdom (B.1.1.7 lineage) is spreading globally and has now acquired an E484K substitution, which confers resistance to certain monoclonal antibodies.

We and our colleagues reported that BNT162b2, a messenger RNA vaccine that expresses the prefusion stabilized full spike glycoprotein (S) of SARS-CoV-2 isolate Wuhan-Hu-1 (GenBank accession number, MN908947.3), is 95% effective against coronavirus disease 2019 (Covid-19).[1] In addition, we reported that recombinant SARS-CoV-2 bearing *S* genes from the B.1.1.7 variant, the variant first identified in South Africa (B.1.351 lineage), and the variant first identified in Brazil (P.1 lineage) remained susceptible to BNT162b2 vaccine–elicited serum neutralization, although at a reduced level for the B.1.351 variant.[2]

To determine whether variants that have emerged more recently are also susceptible to BNT162b2-elicited neutralization, we engineered the complete *S* genes of the variant viruses into the genetic background of USA-WA1/2020 (isolated in January 2020) (Fig. S1 in the Supplementary Appendix, available with the full text of this letter at NEJM.org), which resulted in three recombinant viruses: one with the B.1.429 *S* gene (B.1.429-spike–S13I, W152C, L452R, and D614G), a second with the B.1.526 *S* gene (B.1.526-spike–L5F, T95I, D253G, E484K, D614G, and A701V), and a third with the B.1.1.7 *S* gene plus the E484K substitution (B.1.1.7-spike+E484K–Δ69-70, Δ145, E484K, N501Y, A570D, D614G, P681H, T716I, S982A, and D1118H). All the recombinant viruses produced infectious viral titers of more than 10^7 plaque-forming units (PFUs) per milliliter. The B.1.1.7-spike+E484K virus formed smaller plaques than the other viruses (Fig. S2). All the viruses had similar viral RNA genome to PFU ratios (Fig. S3), which suggests equivalent specific infectivities of the viral stocks.

All the recombinant viruses were analyzed by means of 50% plaque reduction neutralization testing with 20 human serum samples, collected from 15 persons 2 or 4 weeks after the second dose of 30 μg of BNT162b2, which was administered 3 weeks after the first immunization[2] (Fig. S4). All the serum samples neutralized USA-WA1/2020 and the variant viruses at titers of 1:80 or higher. The geometric mean neutralizing titers against USA-WA1/2020, B.1.429-spike, B.1.526-spike, and B.1.1.7-spike+E484K viruses were 520, 394, 469, and 597, respectively (Fig. 1 and Table S1). Thus, as compared with neutralization of USA-WA1/2020, neutralization of B.1.1.7-spike+E484K and B.1.526-spike viruses was approximately equivalent, and neutralization of B.1.429-spike was slightly lower, possibly reflecting the influence of the L452R mutation, which appears to be under positive selective pressure.[3] Our results suggest that, as compared with the previously reported neutralization of B.1.1.7-spike, the additional E484K mutation, which is also found in the B.1.351 and B.1.526 lineages, caused little compromise to neutralization.[4]

An inherent limitation of the study is that new SARS-CoV-2 variants continuously emerge, so the set of strains of current concern constantly shifts. Nevertheless, some mutations are of particular interest. For example, the E484K mutation has arisen convergently, multiple times, in several variants. A second limitation is the potential for mutations to alter neutralization by affecting spike function rather than antigenicity, despite the similar titers and specific infectivities of the viral variant preparations. A third limitation is that BNT162b2 elicits multiple immune effectors, including SARS-CoV-2 spike-specific CD4+ and CD8+ T cells and nonneutralizing antibodies that mediate antibody-dependent cytotoxicity.[4,5] Thus, studies of virus neutraliza-

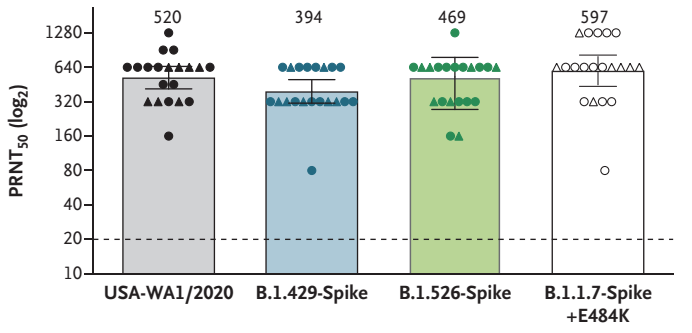

Figure 1. Serum Neutralization of New Variant Strains of SARS-CoV-2 after Two Doses of BNT162b2 Vaccine.

Shown are the results of 50% plaque reduction neutralization testing ($PRNT_{50}$) with the use of 20 samples obtained from 15 trial participants at 2 weeks (circles) or 4 weeks (triangles) after the administration of the second dose of the BNT162b2 vaccine. The mutant viruses were produced by engineering the complete *S* genes from the B.1.429 variant (B.1.429-spike), B.1.526 variant (B.1.526-spike), or B.1.1.7 variant plus an additional E484K mutation (B.1.1.7-spike+E484K) into USA-WA1/2020. Each data point represents the geometric mean $PRNT_{50}$ obtained with a serum sample against the indicated virus, including data from repeat experiments, as detailed in Table S1 in the Supplementary Appendix. The data for USA-WA1/2020 are from two experiments; the data for B.1.429-spike, B.1.526-spike, and B.1.1.7-spike+E484K viruses are from one experiment each. In each experiment, the neutralization titer was determined in duplicate assays, and the geometric mean was calculated. The heights of bars and the numbers over the bars indicate geometric mean titers. The I bars indicate 95% confidence intervals. The dashed line indicates the limit of detection. Statistical analysis was performed with the use of the Wilcoxon matched-pairs signed-rank test. The statistical significance of the difference between geometric mean titers in the USA-WA1/2020 neutralization assay and in each variant virus neutralization assay with the same serum samples are as follows: P=0.002 for B.1.429-spike; P=0.47 for B.1.526-spike; and P=0.04 for B.1.1.7-spike+E484K.

tion by postimmunization serum can show that a variant remains susceptible to one potential mechanism of vaccine-mediated protection but cannot rule out susceptibility to other mechanisms of protection and cannot substitute for clinical evidence of vaccine-mediated protection or escape from that protection.

Because these data show that the newly emerged B.1.526, B.1.429, and B.1.1.7+E484K variants remain susceptible to an important vaccine-elicited immune effector (neutralizing antibody), they confirm the importance of mass immunization with current, highly effective, authorized vaccines as a central strategy to end the Covid-19 pandemic.

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1. Polack FP, Thomas SJ, Kitchin N, et al. Safety and efficacy of the BNT162b2 mRNA Covid-19 vaccine. N Engl J Med 2020;383:2603-15.
2. Liu Y, Liu J, Xia H, et al. Neutralizing activity of BNT162b2-elicited serum. N Engl J Med 2021;384:1466-8.
3. Tchesnokova V, Kulakesara H, Larson L, et al. Acquisition of the L452R mutation in the ACE2-binding interface of spike protein triggers recent massive expansion of SARS-CoV-2 variants. March 11, 2021 (https://www.biorxiv.org/content/10.1101/2021.02.22.432189v2). preprint.
4. Sahin U, Muik A, Vogler I, et al. BNT162b2 induces SARS-CoV-2-neutralising antibodies and T cells in humans. December 11, 2020 (https://www.medrxiv.org/content/10.1101/2020.12.09.20245175v1). preprint.
5. Tauzin A, Nayrac M, Benlarbi M, et al. A single BNT162b2 mRNA dose elicits antibodies with Fc-mediated effector functions and boost pre-existing humoral and T cell responses. March 18, 2021 (https://www.biorxiv.org/content/10.1101/2021.03.18.435972v1). preprint.

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